From Dysmorphology to Next-Generation Phenotyping

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Society for Birth Defects Research and Prevention’s 2020 Virtual 60th Annual Meeting
Conflict of Interest

• Chief Medical Officer for FDNA, company providing Face2Gene app
  – Past: Travel reimbursement and consulting fee
  – Current: Stock options
Dr. Robert L. Brent, MD, PhD

- World renowned expert on embryonal and fetal radiation exposure
- National and international awards for contributions to the field
- Chairman, Department of Pediatrics at Nemours, Jefferson >30 years
Clinical Genetics: Integration of genetic and phenotypic information

Next-Generation Sequencing (NGS)

Next-Generation Phenotyping (NGP)

Image from FDNA
A New Syndrome: Mental Subnormality and Nasal Papillomata

J. M. COSTELLO

Department of Paediatrics, University of Auckland


Figure 1: Case 1, age 41 years. Curly hair, depressed nasal bridge, epicanthic folds.

Figure 2: Nasal papillomata. Case 1.

Figure 3: Case 2, age 4 years. Curly hair, depressed nasal bridge, epicanthic folds.
A New Syndrome: Mental Subnormality and Nasal Papillomata

J. M. COSTELLO
Department of Paediatrics, University of Auckland

Costello, J. M. (1977). Aust. pediat. J., 13, 114-118. A New Syndrome: Mental subnormality and nasal papillomata. Two unrelated children with poor postnatal growth, mental subnormality, similar physical appearance and nasal papillomata present a syndrome for which no cause has been found.

The author is not aware of any comparable cases reported either before or after these children were described at a meeting in 1971 (Costello, 1971). These children are, therefore, presented as a new syndrome of unknown cause.

<table>
<thead>
<tr>
<th>TABLE I</th>
</tr>
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<tbody>
<tr>
<td></td>
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<tr>
<td>Gestation</td>
</tr>
<tr>
<td>Hydramnios</td>
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<tr>
<td>Birth weight</td>
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<tr>
<td>Poor sucking</td>
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<td>Poor postnatal growth</td>
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<tr>
<td>Large head</td>
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<tr>
<td>Short neck</td>
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<tr>
<td>Curly hair</td>
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<tr>
<td>Low set ears</td>
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<tr>
<td>Large ear lobes</td>
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<tr>
<td>Depressed nasal bridge</td>
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<tr>
<td>Nasal papillomata</td>
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<tr>
<td>Strabismus</td>
</tr>
<tr>
<td>Epicanthic folds</td>
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<tr>
<td>Enamel dysplasia</td>
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<tr>
<td>Thick lips</td>
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<tr>
<td>Barrel chest</td>
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<tr>
<td>Short flat hyper-extensible fingers</td>
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<tr>
<td>Thin nails</td>
</tr>
<tr>
<td>Leg abnormalities</td>
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<tr>
<td>Loose Integument hands and feet</td>
</tr>
<tr>
<td>High arched palate</td>
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<tr>
<td>Skin colour</td>
</tr>
<tr>
<td>Increased carrying angle elbow</td>
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<tr>
<td>Mental Subnormality</td>
</tr>
</tbody>
</table>
Costello syndrome

Der Kaloustian et al.,

- 3rd patient reported
- Named Costello syndrome
- Noted similarities to Noonan and cardio-facio-cutaneous (CFC)
Costello syndrome

JM Costello, Am J Med Genet 1996: Costello syndrome: Update on the original cases and commentary

TABLE I. Those Manifestations Frequently Seen in Costello Syndrome and Also Frequently Seen in Noonan and/or CFC Syndromes

<table>
<thead>
<tr>
<th></th>
<th>Costello Syndrome (out of 16 cases)</th>
<th>Noonan syndrome</th>
<th>CPC syndrome</th>
</tr>
</thead>
<tbody>
<tr>
<td>Polyhydramnios</td>
<td>6 (38%)</td>
<td></td>
<td></td>
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<tr>
<td>Postnatal growth deficiency</td>
<td>16 (100%)</td>
<td>27/37 [B]</td>
<td></td>
</tr>
<tr>
<td>Poor suck/poor feeding</td>
<td>15 (94%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mental subnormality</td>
<td>16 (100%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Happy sociable personality</td>
<td>9 (56%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Epiacanthic folds</td>
<td>13 (81%)</td>
<td>12/14 [B]</td>
<td></td>
</tr>
<tr>
<td>Downslanting palpebral fissures</td>
<td>8 (50%)</td>
<td>18/20 [B]</td>
<td></td>
</tr>
<tr>
<td>Strabismus</td>
<td>9 (56%)</td>
<td>18/37? [D]</td>
<td></td>
</tr>
<tr>
<td>High-arched palate</td>
<td>8 (50%)</td>
<td>16/17 [B]</td>
<td></td>
</tr>
<tr>
<td>Depressed nasal bridge</td>
<td>13 (81%)</td>
<td>47% [G]</td>
<td></td>
</tr>
<tr>
<td>Wide or long forehead</td>
<td>6 (38%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Loose skin apart from hands and feet</td>
<td>10 (63%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Curly hair</td>
<td>13 (81%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sparse or short or thin hair</td>
<td>6 (38%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nails thin ± deep set ± other</td>
<td>11 (69)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Short neck</td>
<td>13 (81%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Foot defects</td>
<td>12 (75%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Elbow abnormalities</td>
<td>10 (63%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cardiomyopathy</td>
<td>6 (38%)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Differential diagnosis

Noonan, Costello and Cardio-Facio-Cutaneous (CFC) syndrome

Patient images shown with signed consent
Costello syndrome: Further clinical delineation

Cardiac findings in 63% (N=94) Lin et al., 2002
- 30% cardiovascular malformation (pulmonic valve stenosis)
- 34% hypertrophic cardiomyopathy
- 33% atrial tachycardia (chaotic or multifocal)

Tumor predisposition
- Kerr et al. 1998: Embryonal rhabdomyosarcoma
- Gripp et al. 2002: Increased risk for solid tumors (N=17)
  - Embryonal rhabdomyosarcoma; neuroblastoma; bladder cancer
- Screening protocol
- Kratz et al. 2011: confirmed 15% tumor risk by age 20 years

Cerebellar enlargement and Chiari 1 malformation
- Gripp et al. 2010: 32% Chiari 1 malformation (N=28)
Gene identification: Noonan syndrome

- Linkage to 12q22-qter by Jamieson et al., Nat Genet 1994.
- Heterogeneity noted
- *PTPN11* identified through positional candidate approach by Tartaglia et al., Nat Genet 2001.
- *PTPN11* encodes SHP2, a protein tyrosine phosphatase critical for signal transduction pathways, including the mitogen activated protein kinase (RAS/MAPK) pathway.
Noonan syndrome: KRAS mutations

Growth factor → Cell membrane

- RTK
- CBL
- GRB2
- SHC
- SOS1
- SHOC2
- GAB2
- SHP-2

RAS

active RAS → RAF → MEK → ERK → pERK

Noonan syndrome

Carta et al., AJHG, 2006
Schubbert et al., Nat Genet, 2006

KRAS

PTPN11
Aoki et al., Nature Genetics 2005

HRAS missense mutations in 12/13 patients

- 10 altered Gly12 (7 Ser, 2 Ala, 1 Val)
- 2 Gly13Asp
Gene identification: CFC syndrome

Niihori et al., Nat Genet 2006;
Rodriguez-Viciana et al., Science 2006
– Missense mutations in BRAF, MEK1 or MEK2
• RAS/MAPK pathway activation:
  Shared mechanism for RASopathies
The RAS/MAPK pathway affects cell division and differentiation.
Its primary role is likely for embryologic development.
Early (germline) mutations affect embryologic development and results in syndromic condition.
Secondary role as proto-oncogenes after somatic mutation.

- Delineation from clinical description to molecular cause ~ 30 years
- Numerous genes involved:
  - Testing requires next-generation sequencing approaches
  - Large gene panel or exome analysis
Next-generation sequencing and large databases

Patient 1: Short stature, cryptorchidism, Chiari 1 malformation, macrocephaly, distinctive facial features, slow growing curly hair, developmental delay.
No variant in known RASopathy genes.
\- Exome analysis: de novo \textit{PPP1CB} variant c.146G>C; p.Pro49Arg

Patient 2: Feeding difficulties, short stature, pectus excavatum, developmental delay.
No variant in known RASopathy genes.
\- Exome analysis: de novo \textit{PPP1CB} variant c.166C>G; p.Ala56Pro

GeneMatcher.org
GeneMatcher: 2 additional cases, N=4


OMIM\#617506
Noonan syndrome with loose anagen hair 2 (NSLH2)

Novel syndrome delineation

<table>
<thead>
<tr>
<th>Syndrome</th>
<th>PPP1CB</th>
<th>SHOC2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Short stature</td>
<td>9/15</td>
<td>28/31</td>
</tr>
<tr>
<td>GH deficiency</td>
<td>2/2</td>
<td>14/17</td>
</tr>
<tr>
<td>Macrocephaly (relative)</td>
<td>14/14</td>
<td>29/32</td>
</tr>
<tr>
<td>Cardiac</td>
<td>PVS 2/15 ASD 2/15 VSD 2/15</td>
<td>PVS 10/29 Mitral/ tricuspid 8/24</td>
</tr>
<tr>
<td>HCM</td>
<td>0/15</td>
<td>6/29</td>
</tr>
<tr>
<td>Coarctation/hypoplastic aortic arch</td>
<td>3/15</td>
<td>1 report</td>
</tr>
<tr>
<td>Developmental delay</td>
<td>15/15</td>
<td>5/5</td>
</tr>
<tr>
<td>Learning/performance difficulties</td>
<td>At least 3/4</td>
<td>ADHD common</td>
</tr>
<tr>
<td>Hyperpigmentation</td>
<td>4/5</td>
<td>23/31</td>
</tr>
<tr>
<td>Slow growing hair</td>
<td>8/9</td>
<td>LAH</td>
</tr>
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Computer technology in clinical genetics

Variant identification and analysis
• Next-generation sequencing technology
• Sequencing data analysis through algorithms
• Large databases: gnomAD; ClinVar
• In silico modeling of variant effects on protein product: Mutation Taster, REVEL score
• Matching databases: GeneMatcher

Phenotype analysis
• Human phenotype ontology (HPO) terms
• OMIM database
Next-generation phenotyping

- Next-generation sequencing enabled the identification of novel disease genes and syndromes
- Syndrome identification outpaced the clinical experts’ ability to memorize them
- Syndrome recognition is limited by the clinician’s individual experience and time
- Syndrome diagnosis is hampered by atypical or mild presentations
DeepGestalt is the algorithm used in the Face2Gene app (Face2Gene.com)
- Unconstrained 2D images
- Deep convolutional neural network (DCNN) approach
- Trained on Casia-WebFace dataset for face recognition
- Fine-tuned to >300 syndromes through training on >17,000 validated patient images
- Community driven: Uploaded images are analyzed in a non-identifiable manner, data is used to further train syndrome recognition
DeepGestalt

- Face recognition, pre-processing
- 130 landmarks placed
- Cropped into predefined facial regions, analyzed through DCNN
- Aggregate results read out as syndrome similarity
DeepGestalt


**Multiclass analysis**

Training set: >200 syndromes, trained on >10,000 diagnosed images  
Test set:  
Clinical: 502 patient images of cases submitted and solved over time  
Published: 329 diagnosed patient images from London Medical Database

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<thead>
<tr>
<th></th>
<th>Top 10 accuracy</th>
<th>Top 5 accuracy</th>
<th>Top 1 accuracy</th>
</tr>
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<tbody>
<tr>
<td>Clinical test set</td>
<td>90.6% (CI 88%-93%)</td>
<td>85.4% (CI 82.3%-88.4%)</td>
<td>61.3% (CI 57.2%-65.5%)</td>
</tr>
<tr>
<td>Publication test set</td>
<td>89.4% (CI 86%-92.7%)</td>
<td>83.2% (CI 79%-87.2%)</td>
<td>68.7% (CI 63.52-73.55%)</td>
</tr>
</tbody>
</table>
DeepGestalt in clinic today

Next generation phenotyping through Face2Gene
• Facial photo taken in clinic
• Additional information provided (“short stature” or “intellectual disability”)
• Image and terms analyzed immediately and matching syndromes suggested
• Targeted testing reduces cost, shortens diagnostic odyssey
From DeepGestalt to GestaltMatcher

DeepGestalt

Feature encoder

Classifier

Only trained syndromes

- Noonan
- Coffin Siris 1
- HPMRS
- Kabuki

320-dimensional facial descriptor fine-tuned by 21k photos over 301 syndromes

Not restricted to trained syndromes

Hsieh et al., ESHG meeting June 2020
From DeepGestalt to GestaltMatcher

- Each patient represented by one dot
- The similarity between two patients is quantified by the cosine distance

Hsieh et al., ESHG meeting June 2020
An unusual presentation

Large for gestational age, macrocephaly
Mild gross motor delay
Slow growing hair, lentigines
Seizure
Chiari malformation, syrinx
Hypertrophic cardiomyopathy
HRAS c.186_206dup, p.(Glu62_Arg68dup)

HRAS c.186_206dup, p.(Glu62_Arg68dup)
Gripp et al. Eur J Hum Genet 2020

HRAS c.187_207dup, p.(Glu63_Asp69dup)
Lorenz et al. Hum Mol Genet 2013

HRAS c.187_207dup, p.(Glu63_Asp69dup)

Future use of GestaltMatcher?
Conclusions

• Computer aided sequencing changed genetic testing
• Machine learning will change phenotype analysis
• Together, these technologies will support precision medicine
Acknowledgments

- Patients and families- who allowed me to learn so much!
- Collaborators- national and international- too many to name

Dr. Robert L. Brent, MD, PhD

Thank you!!!
Questions?